REMARKS

This amendment is responsive to the Office Action mailed November 14, 2006. Claims 61-64, 67, 70-73, 83-94, and 104-7 under examination in the present action. Claims 65, 66, 68, 69, 74-82 and 95-103 have been withdrawn. Claims 76, 87 and 104-107 have been cancelled herein.

Applicants acknowledge that the restriction has been made final and that the claims have been restricted to *asd* as the essential gene, which is located extrachromosomally. The claims have been amended accordingly. However, Applicants maintain their traverse of the restriction requirement, particularly to the restriction of particular essential genes as patently distinct inventions. Applicants' application shows that the essential genes are interchangeable in the ELVS system. Applicants expressly reserve their right to petition the restriction requirement and/or pursue all restricted subject matter in divisional applications.

The claims have been amended herein to:

(1) further define the permissive environment as one containing the presence of arabinose and/or having a temperature of over 30°C; and

(2) further define the non-permissive environment as one having a temperature of 30°C or below and/or lacking the presence of arabinose.

Support for the amendments are found throughout the specification. More specifically, support for amendments (1) and (2) above can be found on pages 28-31 (temperature) and 31-33 (arabinose) of the specification.

Additional amendments are discussed below. No new matter is presented.

Introduction

The disclosure, in its broadest sense, relates to environmentally limited viability systems (ELVS) for microbes based on differences in environmental conditions, i.e., permissive and non-permissive environments. Viability of the microorganisms is limited to the permissive environment by specifically expressing one or more genes essential to cell viability only while in the permissive environment, and/or expressing one or more lethal genes only in the non-permissive environment. (*See, e.g.*, page 6, lines 1-12 of the specification.)

Response to issues presented under the doctrine of obvious-type double patenting

In the Office Action, Claims 61-62, 70-71, 73, 84, 88, 91-94, and 104-107 are rejected under the judicially-created doctrine of obviousness-type double patenting as allegedly being unpatentable over Claims 6-9 of U.S. Patent No. 5,840,483 (hereinafter "the '483 patent") and Claims 1, 3, and 7-8 of U.S. Patent No. 5,672,345 (hereinafter "the '345 patent") Specifically, the Examiner contends that although the claims are not identical, they are not patentably distinct, stating:

> "both claim sets are drawn to bacterial cells (Enterobacteriaceae) which lack a functional chromosomal *asd* gene and which contain said gene on an extrachromosomal vector, wherein said cells would be viable in a permissive environment, but non-viable in a non-permissive environment, and wherein the *asd* gene would be expressed in the permissive environment, but not in the non-permissive environment." (Office Action, page 4.)

Applicant traverses. Obvious-type double patenting is a doctrine aimed at preventing an extension of the patent right by seeking additional patents on subject matter that differs insignificantly from a patent already obtained. The doctrine requires rejection of an application claim when the claimed subject matter is not patentably distinct from the subject matter claimed in a commonly owned patent. *In re Braat*, 937 F.2d 589, 592, 19 U.S.P.Q.2D (BNA) 1289, 1291-92 (Fed. Cir. 1991). Its purpose is to prevent an unjustified extension of the term of the right to exclude granted by a patent by allowing a second patent claiming an obvious variant of the same invention to issue to the same owner later. *In re Goodman*, 11 F.3d 1046, 1052, 29 U.S.P.Q.2D (BNA) 2010, 2015 (Fed. Cir. 1993).

The law of obvious-type double patenting was developed to cover the situation where patents are not citable as a reference against each other and therefore can not be examined for compliance with the rule that only one patent is available per invention. Double patenting thus is applied when neither patent is prior art against the other. *General Foods Corp. v. Studiengesellschaft Kohle mbH*, 972 F.2d 1272, 1278-81, 23 U.S.P.Q.2D (BNA) 1839, 1843-46 (Fed. Cir. 1992) (summarizing the criteria for obviousness-type double patenting). As the court explained in *In re Boylan*,

> "it must always be carefully observed that the appellant's patent is not 'prior art' under either section 102 or section 103 of the 1952 Patent Act." 55 C.C.P.A. 1041, 392 F.2d 1017, 1018 n.1, 157 U.S.P.Q. (BNA) 370, 371 n.1 (CCPA 1968).

Analysis

An obvious-type double patenting analysis entails two steps. First, the Examiner must construe the claims in the earlier patent and the claims in the later application and determine the differences. MPEP §804; *Georgia-Pacific Corp. v. United States Gypsum Co.*, 195 F.3d 1322, 1326, 52 U.S.P.Q.2D (BNA) 1590, 1593 (Fed. Cir. 1999). Second, the Examiner must determine whether or not the differences in subject matter between the two claims render the claims patentably distinct. MPEP §804; *Id. at 1327, 52 U.S.P.Q.2D (BNA) at 1595.*

Since the doctrine of double patenting seeks to avoid unjustly extending patent rights at the expense of the public, the focus of any double patenting analysis necessarily is on the claims in the multiple patents or patent applications involved in the analysis. MPEP §804.

The cited patents relate to Applicants' previously disclosed technology known as "balanced-lethal" complementation (i.e., plasmid maintenance), which is described in Applicants' specification:

> "A preferred selection method involves a balanced-lethal host-vector system, where an essential gene is carried on a vector and the chromosomal gene is deleted, creating a balanced-lethal condition. The "lethal" deletion is balanced by the presence of the vector borne copy of the wild-type gene." (Page 12, lines 21-25 of the specification)

The balanced-lethal technology was designed as an alternative plasmid maintenance technique to growing in the presence of antibiotics (with a plasmid-borne copy of an antibiotic resistance gene). A particularly preferred balanced-lethal system involves a non-revertible mutation in the host's native chromosomal *asd* gene, which encodes the enzyme β-aspartate semialdehyde dehydrogenase. β-aspartate semialdehyde dehydrogenase is an essential enzyme for the synthesis of diaminopimelic acid (DAP), required for cell wall/membrane integrity. A copy of the native *asd* gene is then placed on a plasmid, e.g., an expression plasmid. Expression plasmids, depending on their size and/or energy demands, often carry with them selective pressure for not maintaining the plasmid, usually due to the high energy requirements originating from the plasmid. The balanced-lethal technology was developed to counter this selective pressure and to permit the survival only of the population of cells which retain the plasmid.

In contrast to the cited patents, the subject application refers to environmentally activated or repressed control sequences which serve as an "on/off" switch, if you will, controlling the expression of particular genetic components in the ELV system, in this case, the *asd* gene. This regulated, or scheduled, expression of the essential gene can be accomplished, for example, by using various environmentally regulated promoter systems, e.g., the *ara*C-P_{BAD} system (induced in the presence of

arabinose and repressed when arabinose is absent) or the temperature-regulated CI857 repressor system (active as a repressor at temperatures below 30°C).

Applicants note that the cited patent claims all contain the following language or similarly expressed elements:

> "wherein the cells of the strain:
> a) lack a functioning native chromosomal gene encoding beta-aspartate semialdehyde dehydrogenase (Asd);
> b) have present a recombinant gene encoding a functional Asd polypeptide which complements the chromosomal asd mutation, but which cannot replace the defective chromosomal gene by recombination;
> c) have a physical linkage between the recombinant genes encoding the functional Asd polypeptide and the immunogenic antigen, wherein the loss of the recombinant gene encoding the functional Asd polypeptides cause the cells to lyse when the cells are in an environment in which the lack of functional Asd causes the cells to lyse."

However, as discussed above, Applicants point out that the cited claims refer to plasmid maintenance systems which ensure that the antigen-encoding vector is maintained in the vaccine strain population. The present claims utilize an environment-triggered cell death to ensure that the vaccine strain cannot survive in particular environments (e.g., outside the host), as a biological containment system independent of heterologous antigen expression. It is clear that the "balanced-lethal" technology does not and cannot accomplish effective biological containment, because the vaccine cells survive in and out of the host due to the presence of the essential gene on the plasmid. What the balanced-lethal cells *do* ensure is that at any given time the surviving cell population will carry the desired plasmid. While the present invention also contemplates the optional use of the balanced-lethal technology to maintain plasmids, the balanced-lethal systems in no way render obvious a biological containment system that upon transfer of the cell to a pre-selected environment ceases expression of the essential gene.

Therefore, since the methods of the present invention contain components and advantages not taught or suggested by the cited claims of record, the present claims are NOT obvious variants of the prior patented claims, and Applicants respectfully request reconsideration and withdrawal of the rejections under the judicially created doctrine of obvious-type double patenting.

Response to issues presented under 35 U.S.C. §112, first paragraph

In the Office Action, the Examiner has rejected Claims 61-64, 67, 73, 83-89, and 104-107 under 35 U.S.C. §112, first paragraph, as allegedly containing subject matter which was not described in such a

way as to reasonably convey to one skilled in the relevant art that the inventors had possession of the claimed invention.

Specifically, the Examiner objects to the claims because they currently encompass all microbial cells, including bacteria, fungi, and protozoans, many of which are not known to contain the gene *asd*. The Examiner states:

> "Therefore, only bacterial cells containing the claimed environmentally limited viability system meet the written description provision of 35 U.S.C. §112, first paragraph." (Office Action, page 6)

Applicants have amended the claims to specify "bacterial cells". As noted on page 20, lines 7-25 of Applicants specification, the *asd* gene, which encodes β-aspartate semialdehyde dehydrogenase, is required for synthesis of an essential component of the rigid layer of the bacterial cell wall.

Accordingly, Applicants submit that the claims, as amended, are fully described in the specification to reasonably convey to one skilled in the art that the inventor had possession of the claimed invention. Applicants respectfully request reconsideration and withdrawal of the rejection under 35 U.S.C. §112, first paragraph.

Response to issues presented under 35 U.S.C. §112, second paragraph

In the Office Action, the Examiner has rejected Claims 61-64, 67, 70-73, 83-94, and 104-107 under 35 U.S.C. §112, second paragraph, as allegedly being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Specifically, the Examiner objects to Claims 61-64, 67, 70-73, 83-94, and 104-107 because they recite the term "temporarily". While not conceding that this terminology presents any issues under 35 U.S.C. §112, second paragraph, Applicants have deleted the cited phrase from independent Claims 61 and 84 to foster advancement of the preferred embodiments.

Additionally, the Examiner objects to Claims 61, 84, 87-88 because they recite the term "viable". Applicants point out that the claims recite bacterial cells wherein the expression of an essential gene *asd*, which is necessary for the production of the rigid layer of the bacterial cell wall, is limited to permissive environmental conditions. As noted on page 20, lines 7-25 of the specification:

> "Accordingly, a preferred essential gene is *asd*, encoding β-aspartate semialdehyde dehydrogenase, an enzyme required for the synthesis of an essential component of the rigid layer of the bacterial cell wall, namely diaminopimelic acid (DAP). DAP is only synthesized by bacteria and is not prevalent in the environment. DAP is synthesized in six enzymatic steps from β-aspartate semialdehyde, which, in turn, is synthesized in two steps from L-aspartic acid. In the first step, L-aspartic acid is

> phosphorylated by one of several (usually three) β-aspartokinases which are encoded by several (usually three) separate genes regulated independently by repression and/or feedback inhibition of the gene products by the ultimate end products L-threonine, L-methionine, and L-lysine. β-aspartophosphate is converted in one step to β-aspartic semialdehyde by β-aspartic semialdehyde dehydrogenase, the product of the *asd* gene. Mutants with a point mutation in or deletion of the *asd* gene as well as mutants with mutations in any of the six genes specifying the enzymes for converting β-aspartate semialdehyde to DAP have an obligatory requirement for DAP in all media. When DAP-requiring mutants are deprived of DAP, they die and are lysed, releasing their contents."

The definiteness inquiry focuses on whether *those skilled in the art* would understand the scope of the claim *when the claim is read in light of the rest of the specification.* MPEP §2173.02; *Orthokinetics, Inc. v. Safety Travel Chairs, Inc.*, 806 F.2d 1565, 1576, 1 USPQ 2d 1081, 1088 (1986) (emphasis added). "[T]he definiteness of the language must be analyzed--not in a vacuum, but always in light of the teachings of the prior art and of the particular application disclosure as it would be interpreted by one possessing the ordinary level of skill in the pertinent art." *In re Moore, 439 F.2d 1235, 169 USPQ 236 (CCPA 1971).* The court in *In re Moore* further elucidated the above rule of law in a footnote, stating: "It is important here to understand that under this analysis claims which on first reading--in a vacuum, if you will--appear indefinite may *upon a reading of the specification disclosure* or prior art teachings become quite definite." *Moore,* 439 F.2d at 1235, 169 USPQ at 238 (emphasis added).

Applicants submit that the term "viable", particularly in view of the context of the claims and teachings of the specification, would be fully understood by persons skilled in the art. The bacterial cells of the present claims cannot produce diaminopimelic acid (DAP), an essential component of the bacterial cell wall, in the non-permissive environment. A person skilled in the art would fully appreciate that the cells would be viable, that is, possess the essential genes and characteristics for cell survival and propagation, in the permissive environment and would be non-viable, that is, lack essential genes or proteins necessary for cell survival and propagation, in the non-permissive environment.

Therefore, the claims, particularly when viewed in light of the specification, clearly apprises one skilled in the art of its scope and, thereby, serves the notice function required by 35 U.S.C. §112, second paragraph. Nothing more is required of Claims 61, 84, 87-88 under 35 U.S.C. §112, second paragraph.

Last, the Examiner objects to Claims 85 and 89 because the claims include the term "about". Claims 85 and 89 have been amended to delete the term "about". The Claims 85 and 89 have been amended specify that the permissive environment comprises a temperature of greater than 30°C and a

non-permissive environment of less than 30°C. Support for the amendment can be found throughout the specification, *see, e.g.*, page 39, lines 7-29.

In view of the foregoing amendments and remarks, reconsideration and withdrawal of the rejections under 35 U.S.C. §112, second paragraph are requested.

Response to issues presented under 35 U.S.C. §102

Galan et al.

In the Office Action, Claims 61-62, 70-73, 83-84, 87-88, 91-94, and 104-107 are rejected under 35 U.S.C. §102(b) as allegedly anticipated by Galan et al., Gene, 94:29-35 (1990)(hereinafter "Galan").

Specifically, the Examiner states:

> "Galan *et al.* disclose an *asd* mutant (and a method of making said mutant) of *Salmonella* wherein the chromosomal *asd* gene is deleted and the strain contains a plasmid with the *asd* gene (see abstract). The *asd* gene is connected to a P_{trc} promoter, which serves as a means of engineered expression (see page 32, column 32, column 1). The instant specification defines a permissive environment as an environment in which the claimed cells are viable, and a non-permissive environment as one in which the cells are non-viable or temporarily viable. As all gene expression would cease when a cell dies (becomes non-viable), the essential gene (*asd*) of the cells disclosed by Galan *et al.* would not be expressed in a non-permissive environment. Hence, the cells disclosed by Galan *et al.* meet the limitations of the instantly claimed invention." (Office Action, page 11)

A rejection for anticipation under 35 U.S.C. §102(b) requires that each and every feature of the claimed invention be disclosed in a single prior art reference. *See* MPEP §2131. Whereas the references of record fail to disclose or suggest aspects of the invention that are particularly and distinctly claimed, reconsideration and withdrawal of the rejection under 35 U.S.C. §102 are requested.

Applicants disagree with the Examiner's reasoning for rejection and note that it is clear from the specification that the invention relates to live cells with environmentally limited viability and not dead cells, which coincidentally, express no genes and therefore do not express the essential gene *asd* in a non-permissive environment. However, the rejection is moot in view of Applicants amendment to the claims. The claims as amended

(1) further define the permissive environment as one containing the presence of arabinose and/or having a temperature of over 30°C; and

(2) further define the non-permissive environment as one having a temperature of 30°C or below and/or lacking the presence of arabinose.

Galan does not teach environmentally limited viability systems wherein the expression of the essential gene *asd* is limited to a permissive environment containing the presence of arabinose and/or having a temperature of over 30°C and the essential gene is not expressed in a non-permissive environment having a temperature of 30°C or below and/or lacking the presence of arabinose.

Accordingly, because the Galan fails to teach aspects particularly recited in the claims as amended, Galan cannot anticipate Claims 61-62, 70-73, 83-84, 87-88, 91-94, and 104-107 as a matter of law. MPEP §2131. Reconsideration and withdrawal of the rejection under 35 U.S.C. §102(b) are therefore requested.

Curtiss

Similarly, the Examiner rejects Claims 60-62, 70-73, 84, 88, 91-94, and 104-107 under 35 U.S.C. §102(e) as being anticipated by U.S. Patent Nos. 5,840,483 and 5,672,345 (both issued to Curtiss) discussed above. The Examiner's reasoning for rejection is identical to that set forth in the 35 U.S.C. §102(b) rejection over Galan et al.

Applicants comments above apply here as well. Neither of the Curtiss patents teach environmentally limited viability systems wherein the expression of the essential gene *asd* is limited to a permissive environment containing the presence of arabinose and/or having a temperature of over 30°C and the essential gene is not expressed in a non-permissive environment having a temperature of 30°C or below and/or lacking the presence of arabinose.

Accordingly, because the Curtiss patents fail to teach aspects of the claims as amended, they cannot anticipate Claims 61-62, 70-73, 83-84, 87-88, 91-94, and 104-107 as a matter of law. MPEP §2131. Reconsideration and withdrawal of the rejection under 35 U.S.C. §102(e) are requested.

Response to issues presented under 35 U.S.C. §103

Galan and Guzman

In the Office Action, Claims 61-63, 67, 70-73, 83-84, 87-88, 91-94, and 104-107 have been rejected as obvious in view of Galan (*supra*) and Guzman et al., J. Bacteriol., 177:4121-4130 (1995)(hereinafter "Guzman"). The Examiner states:

> "Therefore, it would have been obvious to one of skill in the art to use the *ara*C-Pbad promoter system (as disclosed by Guzman *et al.*) to control expression of the *asd* gene in the bacterial cells of Galan *et al.* because it is useful to use such a promoter to control expression in the study of depletion phenotypes." (Office Action, page 17).

Applicants disagree. Galan has been discussed above. Galan merely teaches the use of "balanced-lethal" technology, which is an alternative plasmid maintenance technique to growing in the presence of antibiotics (with a plasmid-borne copy of an antibiotic resistance gene). The *asd* gene, essential for the viability of the cell, is deleted from the chromosome and a cloned copy is inserted into an expression vector to selectively maintain the plasmid. There is no teaching or suggestion in Galan to regulate the expression of the essential gene *asd*. In fact, in Galan, the *asd* gene is constitutively expressed in all cell environments.

The teachings of Guzman do not cure the deficiencies of Galan. Guzman teaches that the *ara*C-Pbad promoter system has tight regulation, that genes under the control of pBAD can be repressed from a 200- to 1,200 fold reduction . Guzman further teaches that this system is useful in studies to assess the effect of the expression or depletion of the gene product in mutants lacking the particular chromosomal gene.

However, there is no teaching or suggestion in either Galan or Guzman to create an environmentally limited viability system wherein a cell is viable in one environment and not viable in another. As noted in Applicants' specification, this discovery is extremely useful for vaccine microorganisms, where oral administration of live avirulent vaccine microorganisms can lead to fecal shedding of the recombinant microorganisms, with the potential risk that the bacterial vaccine strain will proliferate in nature and infect individuals not selected for immunization.

MPEP §2143.03 states that "[t]o establish *prima facie* obviousness of a claimed invention, all claim limitations must be taught or suggested by the prior art." (emphasis added)

Accordingly, because neither Galan nor Guzman, alone or in combination, teach or suggest controlling the viability of a bacterial cell to permissive and non-permissive environments, the claims cannot be obvious as a matter of law. Reconsideration and removal of the rejection of Claims 61-63, 67, 70-73, 83-84, 87-88, 91-94, and 104-107 are requested.

Galan and Glick

In the Office Action, Claims 61-62, 64, 70-73, 83-86, 87-94, and 104-107 have been rejected as obvious in view of Galan (*see, supra*) and Glick et al., Molecular Biotechnology, Principles and Applications of Recombinant DNA, (1994) ASM press, pp. 90-92) (hereinafter "Glick"). The Examiner states:

> "Therefore, it would have been obvious to one of skill in the art to use the temperature regulated *p*L promoter system (as disclosed by Glick *et al.*) to control expression of the *asd* gene in the bacterial cells of Galan *et al.* because regulatable strong promoters are advantageous to avoid a

> high level of continual expression of a cloned gene which is often detrimental to the host cell." (Office Action, page 19).

Once again, for the same reasons as discussed above, Applicants disagree. The deficiencies of Galan has been previously discussed. Galan merely teaches the use of "balanced-lethal" technology, which is an alternative plasmid maintenance technique to growing in the presence of antibiotics (with a plasmid-borne copy of an antibiotic resistance gene). There is no teaching or suggestion in Galan to regulate the expression of the essential gene *asd*. In fact, in Galan, the *asd* gene is constitutively expressed in all cell environments.

The teachings of Glick do not cure the deficiencies of Galan. Glick teaches the CI857/pL promoter system, noting that this system can be used for temperature-regulated transcription of genes operably linked to the pL promoter.

However, there is no teaching or suggestion in either Galan or Glick (or their combination) to create an environmentally limited viability system wherein a cell is viable in one environment and not viable in another. As noted in Applicants' specification, this discovery is extremely useful for vaccine microorganisms, where oral administration of live avirulent vaccine microorganisms can lead to fecal shedding of the microorganisms, with the potential risk that the bacterial vaccine strain will proliferate in nature and infect individuals not selected for immunization.

MPEP §2143.03 states that "[t]o establish *prima facie* obviousness of a claimed invention, all claim limitations must be taught or suggested by the prior art." (emphasis added)

Accordingly, because neither Galan nor Glick, alone or in combination, teach or suggest controlling the viability of a bacterial cell to permissive and non-permissive environments, the claims cannot be obvious as a matter of law. Reconsideration and removal of the rejection of Claims 61-63, 67, 70-73, 83-84, 87-88, 91-94, and 104-107 are requested.

Curtiss '483, Guzman, Curtiss '345, and Glick

In the Office Action, Claims 61-62, 64, 70-73, 83-86, 87-94, and 104-107 have additionally been rejected as obvious in view various combination of Curtiss '483, Guzman, Curtiss '345 and Glick. The deficiencies of all these references with respect to the present invention have been addressed above. As noted previously, both Curtiss references refer to plasmid maintenance systems which ensure that an antigen-encoding vector is maintained in the vaccine strain population. The present claims utilize an environment-triggered cell death to ensure that a recombinant vaccine strain cannot survive in particular environments (e.g., outside the vaccinated host), thus achieving a biological containment system. It is clear that the "balanced-lethal" technology does not and cannot accomplish effective biological

containment, because the vaccine cells survive in and out of the host due to the presence of the essential gene on the plasmid.

There is no teaching or suggestion in any of Curtiss '483, Guzman, Curtiss '345, or Glick to create an environmentally limited viability system wherein a cell is viable in one environment and not viable in another.

MPEP §2143.03 states that "[t]o establish *prima facie* obviousness of a claimed invention, all claim limitations must be taught or suggested by the prior art." (emphasis added)

Accordingly, because none of the citations, alone or in combination, teach or suggest controlling the viability of a bacterial cell to permissive and non-permissive environments, the claims cannot be obvious as a matter of law. Reconsideration and removal of the rejection of Claims 61-63, 67, 70-73, 83-84, 87-88, 91-94, and 104-107 are requested.

Respectfully submitted,

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